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Declaration under Rule 4.17:

— of inventorship (Rule 4.17(iv)) for US only

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For two-letter codes and other abbreviations, refer to the "Guid-
ance Notes on Codes and Abbreviations" appearing at the begin-
ning of each regular issue of the PCT Gazette.

(54) Title: INTEGRASE COFACTOR

(57) **Abstract:** In a study of HIV-1 integrase (IN) complexes derived from nuclei of human cells stably expressing the viral protein from a synthetic gene it was demonstrated that in the nuclear extracts IN exists as part of a large distinct complex with apparent Stokes radius of 61 Å, which dissociates upon dilution yielding a core molecule of 41 Å. The IN complexes were isolated from cells expressing FLAG-tagged IN. By present invention it was demonstrated that the 41 Å core is tetramer of IN, whereas 61 Å molecules are composed of IN tetramers associated with a cellular protein with an apparent molecular weight of 76 kDa. This integrase interacting protein (Inip76) was found to be identical to LEDGF/DFS70/p75 a protein implicated in regulation of gene expression and cellular stress-response. HIV-1 IN and Inip76 co-localized in the nuclei of human cells stably expressing IN. Furthermore, it has been demonstrated by present invention that recombinant Inip76 strongly promoted strand-transfer activity of HIV-1 IN in vitro. Our findings reveal that the minimal IN molecule in human cells is a tetramer and clearly demonstrates that Inip76 plays a role in retroviral integration. Therefore the present invention provides integrase interacting proteins and more particularly cofactors which promote strand transfer activity of viral integrase, more particularly HIV integrase, and methods and uses relating thereto. The present invention relates to a cellular protein that associates with integrase (integrase interacting protein-Inip), to molecules interacting with Inip and their use as an antiviral. The present invention also relates to antibodies, RNA interference, antigen therapy, gene silencing or antisense inhibition of said integrase interacting protein. The novel integrase interaction protein is a target for HIV replication prevention or inhibition.



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INTERNATIONAL SEARCH REPORT

International Application No

PCT/BE 03/00164

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12N15/11 C07K14/16 C07K14/47 C12N15/62 C12Q1/68
 G01N33/50 A61K38/16 A61K38/17 A61K48/00

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12N C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS, EMBASE, WPI Data, MEDLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>LEH H ET AL: "Determinants of Mg2+-dependent activities of recombinant human immunodeficiency virus type 1 integrase." BIOCHEMISTRY. UNITED STATES 8 AUG 2000, vol. 39, no. 31, 8 August 2000 (2000-08-08), pages 9285-9294, XP001181843 ISSN: 0006-2960 page 9285 -page 9294</p> <p style="text-align: center;">--- -/--</p>	27,28



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

* Special categories of cited documents :

A document defining the general state of the art which is not considered to be of particular relevance

E earlier document but published on or after the international filing date

L document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

O document referring to an oral disclosure, use, exhibition or other means

P document published prior to the international filing date but later than the priority date claimed

T later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

X document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

Y document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

Z document member of the same patent family

Date of the actual completion of the international search

9 June 2004

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INTERNATIONAL SEARCH REPORT

ational Application No

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>PODTELEZHNIKOV ALEXEI A ET AL: "Modeling HIV-1 integrase complexes based on their hydrodynamic properties." BIOPOLYMERS. UNITED STATES JAN 2003, vol. 68, no. 1, January 2003 (2003-01), pages 110-120, XP001181844 ISSN: 0006-3525 page 110-120</p> <p>---</p>	27, 28
A	<p>WO 99/05278 A (BRIGHAM & WOMENS HOSPITAL) 4 February 1999 (1999-02-04)</p> <p>---</p>	
A	<p>WO 00/29578 A (US HEALTH ;GE HUI (US)) 25 May 2000 (2000-05-25)</p> <p>---</p>	
A	<p>OCHS R L ET AL: "AUTOANTIBODIES TO DFS 70 KD/TRANSCRIPTION COACTIVATOR P75 IN ATOPICDERMATITIS AND OTHER CONDITIONS" JOURNAL OF ALLERGY AND CLINICAL IMMUNOLOGY, MOSBY - YEARLY BOOK, INC, US, vol. 105, May 2000 (2000-05), pages 1211-1220, XP002944618 ISSN: 0091-6749</p> <p>---</p>	
A	<p>WO 95/31904 A (UNIV COLUMBIA ;GOFF STEPHEN P (US); KALPANA GANJAM V (US)) 30 November 1995 (1995-11-30)</p> <p>---</p>	
A	<p>DIETZ FRANK ET AL: "The family of hepatoma-derived growth factor proteins: Characterization of a new member HRP-4 and classification of its subfamilies" BIOCHEMICAL JOURNAL, vol. 366, no. 2, 1 September 2002 (2002-09-01), pages 491-500, XP002283776 ISSN: 0264-6021</p> <p>---</p>	
P, X	<p>CHEREPANOV PETER ET AL: "HIV-1 integrase forms stable tetramers and associates with LEDGF/p75 protein in human cells." JOURNAL OF BIOLOGICAL CHEMISTRY, vol. 278, no. 1, 3 January 2003 (2003-01-03), pages 372-381, XP002283777 ISSN: 0021-9258 page 372-381</p> <p>-----</p>	1-29

INTERNATIONAL SEARCH REPORT

International application No.
PCT/BE 03/00164

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
see FURTHER INFORMATION sheet PCT/ISA/210
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.1

Although claim 26 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

Although claim 28 are directed to a diagnostic method practised on the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/BE 03/00164

Patent document cited in search report		Publication date		Patent family member(s)		Publication date
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			EP	1066382 A1		10-01-2001
			WO	9905278 A1		04-02-1999
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			AU	2651495 A		18-12-1995
			WO	9531904 A1		30-11-1995
			US	5872213 A		16-02-1999